



Aalborg Universitet

AALBORG UNIVERSITY
DENMARK

ADM1-based modeling of anaerobic digestion of swine manure fibers pretreated with aqueous ammonia soaking

Jurado, Esperanza; Gavala, Hariklia N.; Skiadas, Ioannis

Published in:

Proceedings of the 4th International Symposium on Energy from biomass and Waste, San Servolo, Venice (Italy), November 12-15

Publication date:

2012

Document Version

Early version, also known as pre-print

[Link to publication from Aalborg University](#)

Citation for published version (APA):

Jurado, E., Gavala, H. N., & Skiadas, I. (2012). ADM1-based modeling of anaerobic digestion of swine manure fibers pretreated with aqueous ammonia soaking. In *Proceedings of the 4th International Symposium on Energy from biomass and Waste, San Servolo, Venice (Italy), November 12-15* IWWG-International Waste Working Group, Hamburg University of Technology.

General rights

Copyright and moral rights for the publications made accessible in the public portal are retained by the authors and/or other copyright owners and it is a condition of accessing publications that users recognise and abide by the legal requirements associated with these rights.

- Users may download and print one copy of any publication from the public portal for the purpose of private study or research.
- You may not further distribute the material or use it for any profit-making activity or commercial gain
- You may freely distribute the URL identifying the publication in the public portal -

Take down policy

If you believe that this document breaches copyright please contact us at vbn@aub.aau.dk providing details, and we will remove access to the work immediately and investigate your claim.

ADM1-BASED MODELING OF ANAEROBIC DIGESTION OF SWINE MANURE FIBERS PRETREATED WITH AQUEOUS AMMONIA SOAKING

E. JURADO*, G. ANTONOPOULOU**, G. LYBERATOS***°, H.N. GAVALA* AND I.V. SKIADAS*

** Aalborg University Copenhagen (AAU-Cph), Department of Biotechnology, Chemistry and Environmental Engineering, Lautrupvang 15, DK 2750 Ballerup, Denmark*

*** Institute of Chemical Engineering and High Temperature Chemical Processes, GR 26504 Patras, Greece*

° School of Chemical Engineering, National Technical University of Athens, GR 15780 Athens, Greece

SUMMARY: Anaerobic digestion of manure fibers present challenges due to their low biodegradability. Aqueous ammonia soaking (AAS) and subsequent ammonia removal has been tested as a simple and cheap method to disrupt the lignocellulose and increase the methane potential and the biogas productivity of manure fibers. In the present study, mesophilic anaerobic digestion of AAS pretreated manure fibers was tested in CSTR-type digesters fed with swine manure and/or a mixture of swine manure and AAS pretreated manure fibers. The Anaerobic Digestion Model No.1 (ADM1) was used for the prediction of the effect that the AAS had on the efficiency of the anaerobic digestion of manure. Kinetic parameters were estimated by fitting of the model to data from manure fed digesters. The model was able to satisfactorily simulate the behaviour of digesters fed with manure. However, the model predictions were poorer for digesters fed with mixture of manure and AAS pretreated fibers, mainly due to the higher hydrolysis rate of the solids of pretreated fibers. Proper modification of the kinetic parameters of ADM1 with emphasis on hydrolysis constants is necessary.

1. INTRODUCTION

Methane production from a variety of biological wastes through anaerobic digestion technology is considered ideal in many ways because of its economic and environmental benefits (Chandra et al., 2012). Therefore, biogas production and utilization has become a major part of the rapidly growing renewable energy sector (Deublein and Steinhauser, 2008).

Denmark is one of the largest producers of pig meat and the use of swine manure in biogas production is a common practice. However, biogas plants digesting liquid manure alone are not economically viable due to the relatively low organic content of the manure. Therefore, current

biogas production in Denmark is based on at least 75% animal manure and up to 25% other (additional) biomasses characterized by high methane potential, such as slaughterhouse wastes, glycerine, crops, animal fat, fish oil, etc. Addition of this type of biomasses increases the methane efficiency and thus the process profitability (Deublein and Steinhauser, 2008). However, due to the increased demand for biomass feedstock in the bioenergy sector, supply of alternative organic fractions from industrial and agricultural sectors for production of biogas is becoming increasingly limited. The methane productivity of swine manure based biogas plants could be increased by applying anaerobic digestion only to the solid fraction of manure (alternatively called manure fibers) which contains most of the organic material. However, the intact rigid lignocellulosic structure of the fibers makes anaerobic digestion process slow and economically unfavourable. Therefore, pretreatment of the solid fraction is a prerequisite for increasing its digestibility (Jurado et al., 2010).

Aqueous Ammonia Soaking (AAS) has been so far tested for bioethanol and chemicals production with satisfactory results (Kim and Lee, 2005; Kim et al., 2008; Ko et al., 2009; Fu and Holtzapple, 2010; Gupta and Lee, 2010; Kim et al., 2010). In comparison with high temperature ammonia pretreatment, AAS is characterized by low energy input, no formation of toxic compounds and no loss of sugars. However, studies on the effect the AAS has on methane production from various biomasses are scarce (Himmelsbach et al., 2010; Jurado et al., 2011, 2012). Jurado et al. (2011, 2012) applied AAS to both raw and digested (before and after anaerobic digestion) swine manure fibers showing an impressive methane potential increase (up to 180% increase). It has to be emphasized that apart the increased methane yield, the ammonia used for the pretreatment can be easily recycled in a full-scale plant resulting in actually no chemicals consumption. Therefore, application of AAS on manure fibers in biogas plants already equipped with ammonia removal infrastructure will constitute a cost-efficient and sustainable pretreatment (or post-treatment) option.

Anaerobic digestion is a complicate and sensitive biological process. Combined with the relatively very long retention times, the experimental determination of the values of the operational parameters that will maximize the process efficiency may be prohibitively time consuming and expensive. The development and use of mathematical models able to predict and describe the behaviour of experimental systems allows for a safe design and scale-up of anaerobic digestion units without the need for time consuming experimental tests. In order to describe the performance and kinetics of the anaerobic digestion process for methane generation, several anaerobic digestion models were developed during the last 30 years (Gavala et al., 2003). The latest developed model is the International Water Association (IWA) Anaerobic Digestion Model No. 1 (ADM1), published in 2002 (Batstone et al., 2002). Although ADM1 was principally developed for anaerobic digestion of sludge, its structure allows modeling of anaerobic treatment of different effluents (Batstone and Keller, 2003; Ersahin et al., 2007) like (agro-)industrial wastewaters (Rajinikanth et al., 2008; Kalfas et al., 2006; Dereli et al., 2010; Antonopoulou et al., 2012), manure and energy crops (Lubken et al., 2007; Rojas et al., 2011; Zhou et al., 2011), manure and organic wastes (Schon, 2009) and manure alone (Gali et al., 2009). All these studies show the increasing trend of applications of ADM1 model as well as the necessity of appropriate modifications in its structure in order to simulate different anaerobic digestion concepts.

In the present study, anaerobic digestion of AAS pretreated manure fibers has been applied in mesophilic CSTR-type digesters. Subsequently, Anaerobic Digestion Model No1 (ADM1) was used to simulate the anaerobic digestion process in the digesters and to assess the effect that the addition of pretreated fibers had on the kinetics of the process.

2. MATERIALS AND METHODS

2.1 Analytical methods

Determination of total solids (TS) and volatile solids (VS) was carried out according to APHA (2005). Total and soluble Chemical Oxygen Demand (COD) were measured with Hach Lange kits LCK-914 and LCK-514 respectively. Ammonium nitrogen ($\text{NH}_4\text{-N}$) analysis was performed with Hach Lange kit LCK-305. Phosphorus analysis was carried out by applying persulphate digestion and subsequent ascorbic acid photometric determination according to APHA (2005). Kjeldahl nitrogen determination was made using the micro-Kjeldahl method for the digestion followed by distillation and finally titration with 0.02N H_2SO_4 (APHA, 2005). Detection and quantification of sugar monomers (glucose, xylose and arabinose) was made with HPLC-RI equipped with an Aminex HPX-87H column (BioRad) at 60°C. Two groups of carbohydrates were determined in the samples of raw and pretreated manure fibers: total carbohydrates, including those bound in the lignocellulosic biomass and simple sugars (Haagensen et al., 2009). Analysis of the two groups of carbohydrates was carried out based on the NREL analytical procedures (Sluite et al., 2011). Biogas composition in methane was measured with a gas chromatograph (SRI GC model 310) equipped with a thermal conductivity detector and a packed column (Porapak-Q, length 6ft and inner diameter 2.1 mm). For the quantification of VFA, 1 ml of liquid sample was acidified with 17% H_3PO_4 and filtered through minisart high flow filter (pore size 0.5 μm). VFAs were analyzed with a gas chromatograph (PerkinElmer, Clarus 400) with a flame ionization detector and a capillary column (Agilent HP-FFAP, 30 m long, 0.53 mm inner diameter).

2.2 Experimental procedure

2.2.1 Influent characterisation

AAS pretreatment of fibers was performed according to Jurado et al. (2012). Manure and AAS-pretreated raw manure fibers were characterized in terms of total and soluble COD, total carbohydrates and free sugars, total and soluble Kjendahl nitrogen and $\text{NH}_3\text{-N}$, volatile fatty acids (valeric, butyric, propionic and acetic acids), inorganic phosphorus and inorganic carbon. Particulate and soluble inerts were determined as the residual COD after three months of batch anaerobic digestion. Particulate carbohydrates and proteins were calculated as the difference between total and free carbohydrates/sugars and total and soluble Kjendahl nitrogen, respectively. Aminoacids were calculated as the difference between soluble Kjendahl nitrogen and $\text{NH}_3\text{-N}$. Calculation of particulate lipids was based on the difference between non-soluble COD (total – soluble COD) and the sum of particulate carbohydrates, proteins and inerts, while long chain fatty acids were calculated as the difference between soluble COD and all measured soluble components (sugars, aminoacids, volatile fatty acids and soluble inerts).

2.2.1 Continuous experiments

In the present study, anaerobic digestion of AAS pretreated manure fibers in continuous mode has been applied. Two mesophilic (38°C) CSTR-type digesters of 3 L useful volume were operated in parallel. The first was fed with swine manure and the second was initially fed with manure and after 110 days the manure feed was replaced with a mixture of swine manure and AAS pretreated manure fibers (at a ratio of 0.52:0.48 on Total Solids basis) at a hydraulic retention time of 25 d. The feeding was intermittent and repeated once a day.

Daily monitoring of the digesters included biogas production and composition in methane, pH, volatile fatty acids and soluble COD concentration. When the two digesters reached steady

state, complete characterisation was performed in terms of almost all measurable components of ADM1. Subsequently, the digesters were subjected to impulse disturbances of different substrates in order to study the dynamics of the processes. The substrates used for the disturbances were acetic, propionic and butyric acids and the soluble part of the influent of the digesters. The response of the bioreactors to these impulses was monitored mainly through measurements of biogas production and composition in methane, pH, VFA concentration and soluble COD.

2.3 Modelling

Anaerobic Digestion Model No1 (ADM1) was fitted on the data from the continuous anaerobic digestion of manure. Subsequently, the model was used to simulate the anaerobic digestion process in both reactors and thus the ability of the model to predict the effect that the addition of AAS pretreated fibers had on the process efficiency was assessed. The reactors were allowed to reach steady state and were subjected to impulse disturbances of different substrates-intermediates of the anaerobic digestion process (e.g. acetic, propionic and butyric acids) as well as soluble fraction of the feed in order to study the process kinetics. First, the IWA anaerobic digestion model (ADM1) was fitted to the experimental data obtained from the manure-fed digester (reactor 1) using non-linear parameter estimation. The software used was Aquasim 2.1 g and the secant method was applied for optimisation. Parameter estimation focused on the hydrolysis constants of carbohydrates (khydr_ch), proteins (khydr_pr) and lipids (khydr_li) and maximum uptake rates of long chain fatty acids (Km_fa) and volatile fatty acids (km_c4, km_pro and km_ac for butyric, propionic and acetic acid, respectively). The values suggested by Batstone et al. (2002) were used for the remaining kinetic parameters and all stoichiometric coefficients. Subsequently, the model developed on the first, fed with manure, reactor was used to simulate the performance of the second reactor (reactor 2), which was fed with a mixture of manure and AAS pretreated manure fibers.

3. RESULTS AND DISCUSSION

3.1 Influent characterisation

The characteristics of manure and AAS-pretreated raw manure fibers are shown in Table 1. The TS content of manure and fibers was 37 and 129 g/L, respectively. As anticipated, the AAS-pretreated fibers consisted mainly of particulate organic matter with carbohydrates being a substantial fraction while manure was also rich in soluble organic matter, mainly in long chain and volatile fatty acids.

3.2 Continuous experiments

Operating conditions and steady state characteristics of the anaerobic digesters fed with manure and a mixture of AAS-pretreated raw manure fibers are shown in Table 2. Although the operating characteristics were the same for both reactors and despite the fact that manure fibers were characterised by a much lower content in soluble organic material (Table 1) the biogas production of the second reactor was 22% higher than that of the first reactor. The methane yield (Table 2) was calculated as 0.26 L CH₄ / g TS added for reactor 1 at steady state and 0.24 L CH₄ / g TS added for reactor 2 during the second steady state when the digester was fed with mixture of manure and AAS-pretreated fibers. Given that the TS ratio of manure:fibers in the influent of the digester 2 was 0.52:0.48 and assuming that the methane yield due to the manure fraction was

0.26 L CH₄ / g TS also in reactor 2, then the methane yield of AAS-pretreated fibers was calculated to be 0.23 L CH₄ / g TS. In previous studies, the methane yield from swine manure fibers was measured in the range of 0.12-0.13 L CH₄ / g TS in batch experiments (Jurado et al., 2012). Consequently, AAS pretreatment resulted to a significant increase of methane yield of manure fibers.

Values of the kinetic constants obtained through fitting of the experimental data of Reactor 1 (manure-fed reactor) to ADM1 are summarised in Table 3. It is noticeable that hydrolysis of particulate matter was extremely slow, with carbohydrates being non-hydrolyzable. Experimental and model simulated biogas production and composition in methane are shown in Figures 1 and 2, while acetate level throughout the duration of the continuous experiment in reactor 1 as well as during impulse disturbances is shown in Figure 3.

Table 1. Characteristics of manure and AAS-pretreated raw manure fibers

Characteristics, particulate matter	Manure	AAS-pretreated raw fibers
COD, kg / 100 kgTS	79.11	98.6
Carbohydrates, kg COD/ 100 kg TS	6.10	47.79
Proteins, kg COD/ 100 kg TS	28.00	20.12
Lipids, kg COD/ 100 kg TS	32.65	14.58
Inerts, kg COD/ 100 kg TS	12.37	16.08
Characteristics, soluble matter		
COD, kg / 100 kgTS	91.83	8.10
Sugars, kg COD/ 100 kg TS	0	0
Aminoacids, kg COD/ 100 kg TS	18.92	0.21
Long chain fatty acids, kg COD/ 100 kg TS	21.25	3.89
Valeric acid, kg COD/ 100 kg TS	0	0.11
Butyric acid, kg COD/ 100 kg TS	3.98	0.16
Propionic acid, kg COD/ 100 kg TS	8.46	0.34
Acetic acid, kg COD/ 100 kg TS	34.75	2.07
Inerts, kg COD/ 100 kg TS	4.46	1.34
Inorganic carbon, kmole / 100 kg TS	0.53	0.099
Inorganic phosphorus, kmole / 100 kg TS	9.59x10 ⁻³	4.77x10 ⁻³
Inorganic nitrogen (NH ₃ -N), kmole / 100 kg TS	0.73	8.63x10 ⁻³

Table 2. Operating conditions and steady state characteristics of the digesters fed with manure (Reactor 1) and a mixture of manure and AAS-pretreated manure fibers (Reactor 2), respectively.

Operating Conditions	Reactor 1	Reactor 2
Hydraulic Retention Time (d)	25	23
Influent total COD (g /L)	72	70
Influent soluble COD (g/L)	38	28
Characteristics at steady state		
Biogas productivity (L/L/d)	0.86	1.05
Methane (%)	61	61
Methane yield (L/g COD added)	0.18	0.22
Methane yield (L/g TS added)	0.26	0.24
Volatile Fatty Acids (mg/L)	160	170
pH	8.3	8.3
Effluent soluble COD (g/L)	5.4	5

Table 3. Values of the kinetic constants obtained through fitting of the experimental data of Reactor 1 (manure-fed reactor) to ADM1.

Kinetic constant	Value
k_{hydr_ch}, d^{-1}	0
k_{hydr_pr}, d^{-1}	3.0×10^{-3}
k_{hydr_li}, d^{-1}	2.8×10^{-4}
$km_fa, kg\ COD / kg\ COD / d$	0.93
$km_c4, kg\ COD / kg\ COD / d$	13.10
$km_pro, kg\ COD / kg\ COD / d$	6.56
$km_ac, kg\ COD / kg\ COD / d$	45.02

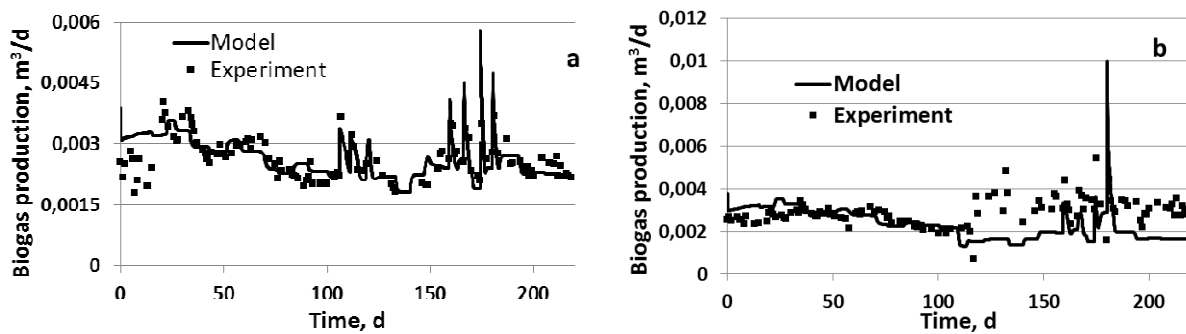


Figure 1. Experimental (■) and model simulated (—) biogas production throughout the duration of the continuous experiment as well as during impulse disturbances in reactor 1 (manure-fed reactor) (a) and in reactor 2 (fed with mixture of manure and AAS pretreated fibers after day 110) (b).

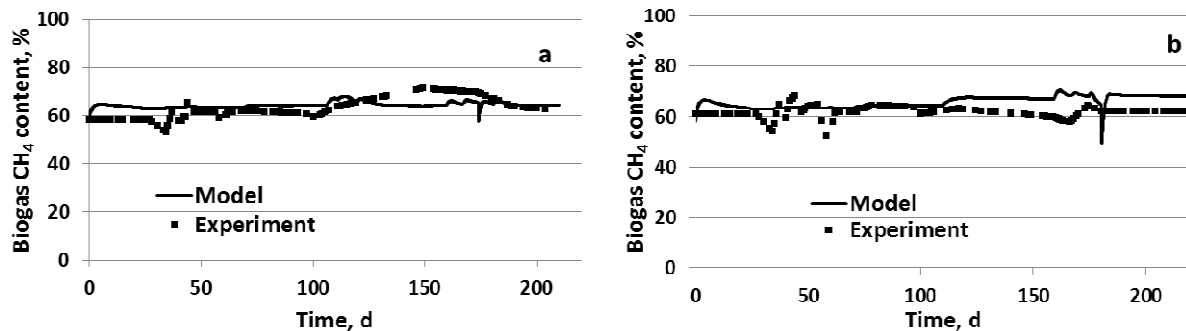


Figure 2. Experimental (■) and model simulated (—) biogas composition in CH₄ throughout the duration of the continuous experiment as well as during impulse disturbances in reactor 1 (manure-fed reactor) (a) and in reactor 2 (fed with mixture of manure and AAS pretreated fibers after day 110) (b).

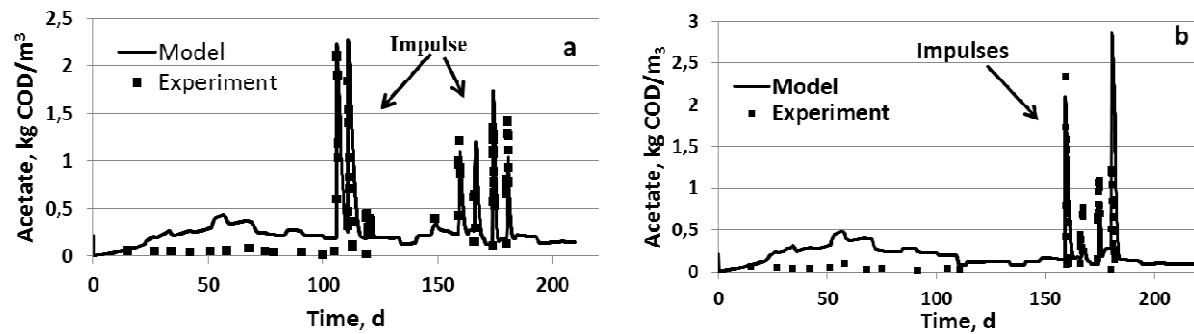


Figure 3. Experimental (■) and model simulated (—) acetate concentration throughout the duration of the continuous experiment as well as during impulse disturbances in reactor 1 (manure-fed reactor) (a) and in reactor 2 (fed with mixture of manure and AAS pretreated fibers after day 110) (b).

As it may be seen from Figures 1a, 2a and 3a the ADM1 was able to satisfactorily simulate the behaviour of digester 1 under steady state and during the impulse disturbances of different substrates-intermediates of the anaerobic digestion process. Also, the model with the kinetic parameters resulted from the fitting to the experimental data of reactor 1 was able to adequately simulate the behaviour of reactor 2 while the reactor was fed only with manure (0-109 days) (Figures 1b, 2b and 3b). In all cases, ADM1 could very well simulate the profiles of volatile fatty acids (results shown in Figures 3a and 3b include only acetate). However, the model with the above determined kinetic parameters could poorly simulate the effect of the addition of AAS pretreated fibers in the influent of reactor 2 after day 110. Specifically, significantly lower biogas production (Figure 1b) and higher methane percentage (Figure 2b) was predicted when reactor 2 was fed with the mixture of manure and fibers. This may be explained by the different lipids and carbohydrates content in the manure compared to AAS pretreated fibers (Table 1). Anaerobic degradation of lipids results in higher methane content compared to the anaerobic degradation of carbohydrates. The model was adapted to the higher lipids content and the lower (and hardly biodegradable) carbohydrates content of the manure and therefore it overestimated the methane content and underestimated the methane production when the reactor was fed with AAS pretreated fibers (containing less lipids and more carbohydrates). The above trend becomes more intense since the AAS-pretreatment releases carbohydrates which are easier biodegradable than the carbohydrates in the manure. This may be explained by a higher hydrolysis rate of the solids of AAS-pretreated fibers and proper modification of the kinetic parameters of ADM1 with emphasis on hydrolysis constants is necessary.

4. CONCLUSIONS

Aqueous Ammonia Soaking of manure fibers increases the methane potential of the fibers and results in more efficient anaerobic digestion of manure. The Anaerobic Digestion Model No.1 was used for the prediction of the effect that the addition of AAS pretreated fibers had on the efficiency of the anaerobic digestion of manure. ADM1 kinetic parameters were estimated by fitting of the model to data from digesters fed with manure. The model was able to satisfactorily simulate the behaviour of digesters fed with manure under steady state and during disturbances of different substrates-intermediates of the anaerobic digestion process. However the model predictions were poorer (the methane content was overestimated and the methane production was underestimated) for digesters fed with mixture of manure and AAS pretreated fibers. This is

due to the higher hydrolysis rate of the solids of pretreated fibers which requires the modification of the kinetic parameters of ADM1 with emphasis on hydrolysis constants.

ACKNOWLEDGEMENTS

The authors wish to thank the EUDP-2008, Energistyrelsen, Copenhagen for the financial support of this work under RETROGAS project.

REFERENCES

- Antonopoulou G., Gavala H.N., Skiadas I.V. and Lyberatos G. (2012). ADM1-based modeling of methane production from acidified sweet sorghum extract in a two stage process. *Bioresour Technol.*, vol. 106, 10-19.
- APHA: Standard Methods for the Examination of Water and Wastewater (2005). 21st edn, American Public Health Association/ American Water Works Association/ Water Pollution Control Federation. Washington DC, USA.
- Batstone D.J. and Keller J. (2003). Industrial application of the IWA Anaerobic Digestion Model No. 1 (ADM1). *Water Sci. Technol.*, vol. 47, n. 12, 199–206.
- Batstone D.J., Keller J, Angelidaki I., Kalyuzhnyi S., Pavlostathis S.G., Rozzi A., Sanders W., Siegrist H. and Vavilin V. (2002). Anaerobic Digestion Model No. 1 (ADM1) IWA Scientific and Technical Report No. 13. IWA Publishing, London.
- Chandra R., Takeuchi H. and Hasegawa T. (2012). Methane production from lignocellulosic agricultural crop wastes: A review in context to second generation of biofuel production. *Renew. Sust. Energ. Rev.*, vol. 16, n. 3, 1462-1476.
- Dereli R.K., Ersahin M.E., Ozgun H., Ozturk I. and Aydin A.F. (2010). Applicability of Anaerobic Digestion Model No. 1 (ADM1) for a specific industrial wastewater: opium alkaloid effluents. *Chem. Eng. J.*, vol. 165, n. 1, 89–94.
- Deublein D and Steinhauser A. (2008). *Biogas from Waste and Renewable Resources*. WILEY-VCH Verlag GmbH & Co. KGaA, Weinheim.
- Ersahin M.E., Insel G., Dereli R.K., Ozturk I. and Kinaci C. (2007). Model based evaluation for the anaerobic treatment of corn processing wastewaters. *Clean Soil Air Water*, vol. 35, n. 6, 576–581.
- Fu Z. and Holtzapple M.T. (2010). Anaerobic mixed-culture fermentation of aqueous ammonia-treated sugarcane bagasse in consolidated bioprocessing. *Biotechnol. Bioeng.*, vol. 106, 216-227.
- Gali A., Benabdallah T., Astals S. and Mata-Alvarez J. (2009). Modified version of ADM1 model for agro-waste application. *Bioresour Technol.*, vol. 100, n. 11, 2783-2790.
- Gavala H.N., Angelidaki I. and Ahring B.K. (2003). Kinetics and modeling of anaerobic digestion process. *Biomethanation I, Advances in Biochemical Engineering/Biotechnology*, Scheper (Ed), Springer-Verlag, vol. 81, 57–93.
- Gupta R. and Lee Y. (2010) Investigation of biomass degradation mechanism in pretreatment of switchgrass by aqueous ammonia and sodium hydroxide. *Bioresour. Technol.*, vol. 101, 8185-8191.
- Haagensen F., Skiadas I.V., Gavala H.N. and Ahring B.K. (2009). Pre-treatment and ethanol fermentation potential of olive pulp at different dry matter concentrations. *Biomass Bioenerg.*,

vol. 33, 1643-1651.

- Himmelsbach J., Raman D., Anex R., Burns R. and Faulhaber C. (2010). Effect of ammonia soaking pretreatment and enzyme addition on biochemical methane potential of switchgrass. *Transactions of the ASABE*, vol. 53, 1921-1927.
- Jurado E., Gavala H.N. and Skiadas I.V. (2012). Application of aqueous ammonia soaking for enhancement of methane potential of swine manure fibers. *Proceedings of the 4th International Conference on Engineering for Waste and Biomass Valorisation*, Porto, Portugal.
- Jurado E., Gavala H.N., Rohold L. and Skiadas I.V. (2010). Cost-effective production of biogas from manure – Retrogas project. *Proceedings of the WasteEng10 - 3rd International Conference on Engineering for Waste and Biomass Valorisation*, Beijing,.
- Jurado E., Skiadas I.V. and Gavala H.N. (2011). Enhanced methane productivity from swine manure fibers by aqueous ammonia soaking pretreatment. *Proceedings of the International Symposium on Anaerobic Digestion of Solid Waste and Energy Crops*. Vienna, Austria.
- Kalfas H., Skiadas I.V., Gavala H.N., Stamatelatos K. and Lyberatos G. (2006). Application of ADM1 for the simulation of anaerobic digestion of olive pulp under mesophilic and thermophilic conditions. *Water Sci. Technol.*, vol. 54, n. 4, 149–156.
- Kim M., Aita G. and Day D.F. (2010). Compositional changes in sugarcane bagasse on low temperature, long-term diluted ammonia treatment. *Appl. Biochem. Biotechnol.*, vol. 161, 34–40.
- Kim T.H. and Lee Y. (2005). Pretreatment of corn stover by soaking in aqueous ammonia. *Appl. Biochem. Biotechnol.*, 121-124, 1119-1131.
- Kim T.H., Taylor F. and Hicks K.B. (2008). Bioethanol production from barley hull using SAA (soaking in aqueous ammonia) pretreatment. *Bioresour. Technol.*, vol. 99, 5694-5702.
- Ko J.K., Bak J.S., Jung M.W., Lee H.J., Choi I.G., Kim T.H. and Kim K.H. (2009). Ethanol production from rice straw using optimized aqueous-ammonia soaking pretreatment and simultaneous saccharification and fermentation processes. *Bioresour. Technol.*, vol. 100, 4374-4380.
- Lubken M., Wichern M., Schlattmann M., Gronauer A. and Horn, H. (2007). Modelling the energy balance of an anaerobic digester fed with cattle manure and renewable energy crops. *Water Res.*, vol. 41, 4085–4096.
- Rajinikanth R., Ramirez I., Steyer J.P., Mehrotra I., Kumar P., Escudie R. and Torrijos M. (2008). Experimental and modeling investigations of a hybrid upflow anaerobic sludge-filter bed (UASFB) reactor. *Water Sci. Technol.*, vol. 58, n. 1, 109–117.
- Rojas C., Uhlenhut F., Schlaak M., Borchert A. and Steinigeweg S. (2011). Simulation of the anaerobic processes for the biogas production. *Chem Ing Technik*, vol. 83, 306–321.
- Schon M. (2009). Numerical Modelling of Anaerobic Digestion Processes in Agricultural Biogas Plants. Ph.D. Thesis, University of Innsbruck, Innsbruck, Austria.
- Sluite A., Hames B., Ruiz R., Scarlata C., Sluiter J., Templeton D. and Crocker D. (2011). Determination of structural Carbohydrates and Lignin in Biomass. National Renewable Energy Laboratory, Available at: www.nrel.gov/biomass/pdfs/42618.pdf (accessed in 2012).
- Zhou H., Löffler D. and Kranert M. (2011). Model-based predictions of anaerobic digestion of agricultural substrates for biogas production. *Bioresour. Technol.*, vol. 102, n. 23, 10819–10828.